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(54) Title: NOVEL ANGIOGENESIS INHIBITORS

(57) Abstract

The present invention relates to compounds which inhibit tyrosine kinase enzymes, compositions which contain tyrosine kinase inhibiting compounds and methods of using tyrosine kinase inhibitors to treat tyrosine kinase–dependent diseases/conditions such as angiogenesis, cancer, atherosclerosis, diabetic retinopathy or autoimmune diseases, in mammals.

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TITLE OF THE INVENTION
NOVEL ANGIOGENESIS INHIBITORS

BACKGROUND OF THE INVENTION

Tyrosine kinases are a class of enzymes that catalyze the transfer of the terminal phosphate of adenosine triphospate to tyrosine residues in protein substrates. Tyrosine kinases are believed, by way of substrate phosphorylation, to play critical roles in signal transduction for a number of cell functions. Though the exact mechanisms of signal transduction is still unclear, tyrosine kinases have been shown to be important contributing factors in cell proliferation, carcinogenesis and cell differentiation. Accordingly, inhibitors of these tyrosine kinases are useful for the prevention and treatment chemotherapy of proliferative diseases dependent on these enzymes.

For example, a method of treatment described herein relates to neoangiogenesis. Neoangiogenesis occurs in conjunction with tumor growth and in certain diseases of the eye. It is characterized by excessive activity of vascular endothelial growth factor.

Vascular endothelial growth factor (VEGF) binds the high affinity membrane-spanning tyrosine kinase receptors KDR and Flt-1. Cell culture and gene knockout experiments indicate that each receptor contributes to different aspects of angiogenesis. KDR mediates the mitogenic function of VEGF whereas Flt-1 appears to modulate non-mitogenic functions such as those associated with cellular adhesion. Inhibiting KDR thus modulates the level of mitogenic VEGF activity.

Vascular growth in the retina leads to visual degeneration culminating in blindness. VEGF accounts for most of the angiogenic activity produced in or near the retina in diabetic retinopathy. Ocular VEGF mRNA and protein are elevated by conditions such as retinal vein occlusion in primates and decreased pO_2 levels in mice that lead to neovascularization. Intraocular

injections of anti-VEGF monoclonal antibodies or VEGF receptor immunofusions inhibit ocular neovascularization in both primate and rodent models. Regardless of the cause of induction of VEGF in human diabetic retinopathy, inhibition of ocular VEGF is useful in treating the disease.

Expression of VEGF is also significantly increased in hypoxic regions of animal and human tumors adjacent to areas of necrosis. Monoclonal anti-VEGF antibodies inhibit the growth of human tumors in nude mice. Although these same tumor cells continue to express VEGF in culture, the antibodies do not diminish their mitotic rate. Thus tumor-derived VEGF does not function as an autocrine mitogenic factor. Therefore, VEGF contributes to tumor growth in vivo by promoting angiogenesis through its paracrine vascular endothelial cell chemotactic and mitogenic activities. These monoclonal antibodies also inhibit the growth of typically less well vascularized human colon cancers in athymic mice and decrease the number of tumors arising from inoculated cells. Viral expression of a VEGF-binding construct of Flk-1, the mouse KDR receptor homologue, truncated to eliminate the cytoplasmic tyrosine kinase domains but retaining a membrane anchor, virtually abolishes the growth of a transplantable glioblastoma in mice presumably by the dominant negative mechanism of heterodimer formation with membrane spanning endothelial cell VEGF receptors. Embryonic stem cells, which normally grow as solid tumors in nude mice, do not produce detectable tumors if both VEGF alleles are knocked out. Taken together, these data indicate the role of VEGF in the growth of solid tumors. Inhibition of KDR or Flt-1 is implicated in pathological neoangiogenesis, and these are useful in the treatment of diseases in which neoangiogenesis is part of the overall pathology, e.g., diabetic retinal vascularization, as well as various forms of cancer.

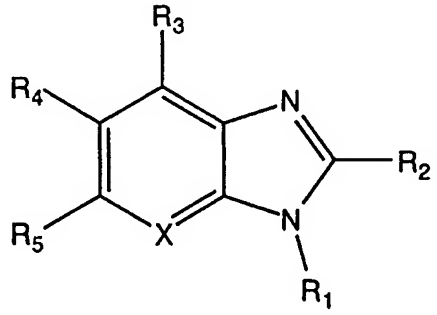
Cancers which are treatable in accordance with the present invention demonstrate high levels of gene and protein

expression. Examples of such cancers include cancers of the brain, genitourinary tract, lymphatic system, stomach, larynx and lung. These include histiocytic lymphoma, lung adenocarcinoma and small cell lung cancers. Additional examples include cancers in which overexpression or activation of Raf-activating oncogenes (e.g., K-ras, erb-B) is observed. More particularly, such cancers include pancreatic and breast carcinoma.

The present invention relates to compounds which inhibit tyrosine kinase enzymes, compositions which contain tyrosine kinase inhibiting compounds and methods of using tyrosine kinase inhibitors to treat tyrosine kinase-dependent diseases/conditions such as neoangiogenesis, cancer, atherosclerosis, diabetic retinopathy or inflammatory diseases, in mammals.

SUMMARY OF THE INVENTION

A compound is disclosed in accordance with formula I:



I

or a pharmaceutically acceptable salt, hydrate or prodrug thereof,

wherein

X is N or C;

R_1 is H, C_{1-10} alkyl, C_{3-6} cycloalkyl, C_{5-10} aryl, halo, OH, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl; said alkyl, alkenyl, alkynyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a;

R_2&R_3 are independently H, C_{1-6} alkyl, C_{5-10} aryl, C3-6 cycloalkyl, OH, NO_2, -NH_2, or halogen;

R_4 is H, C_{1-10} alkyl, C3-6 cycloalkyl, C_{1-6} alkoxy C_{2-10} alkenyl, C_{2-10} alkynyl, C_{5-10} aryl, C_{3-10} heterocyclyl, C_{1-6} alkoxyNR_7R_8, NO_2, OH, -NH_2 or C_{5-10} heteroaryl, said alkyl, alkenyl, alkynyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a;

R_5 is H, or C_{1-6} alkyl, OR, halo, NH_2 or NO_2;

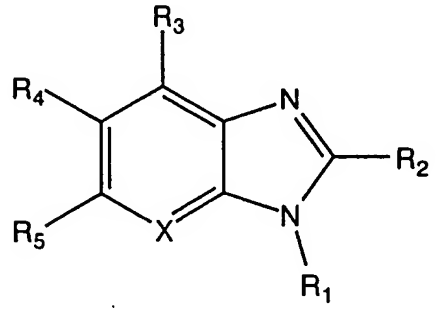
R^a is H, C_{1-10} alkyl, halogen, NO_2, OR, -NR, NR_7R_8, R_7R_8, C_{5-10} aryl, C_{5-10} heteroaryl or C_{3-10} heterocyclyl;

R is H, or C_{1-6} alkyl, C_{1-6} alkylR_9;

R_9 is C_{5-10} aryl, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl; and

R7&R8 are independently H, C_{1-10} alkyl, C3-6 cycloalkyl, COR, C_{5-10} aryl, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl or NR7R8 can be taken together to form a heterocyclic 5-10 membered saturated or unsaturated ring containing, in addition to the nitrogen atom, one to two additional heteroatoms selected from the group consisting of N, O and S.

Also disclosed is a pharmaceutical composition which is comprised of a compound represented by the formula I:



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wherein R_1, R_2, R_3, R_4 and R_5 are described as above or a pharmaceutically acceptable salt or hydrate or prodrug thereof in combination with a carrier.

Also included is a method of treating or preventing a tyrosine kinase dependent disease or condition in a mammal which comprises administering to a mammalian patient in need of such treatment a tyrosine kinase dependent disease or condition treating amount of a compound of formula I or a pharmaceutically acceptable salt, hydrate or pro-drug thereof.

Also included is a method of treating or preventing cancer in a mammalian patient in need of such treatment which is comprised of admininstering to said patient an anti-cancer effective amount of a compound of formula I or a pharmaceutically acceptable salt, hydrate or pro-drug thereof.

Also included in the present invention is a method of treating or preventing diseases in which neoangiogenesis is implicated, which is comprised of administering to a mammalian patient in need of such treatment a compound of formula I or a pharmaceutically acceptable salt, hydrate or pro-drug thereof in an amount which is effective for reducing neoangiogenesis.

More particularly, a method of treating or preventing ocular disease in which neoangiogenesis occurs is included herein, which is comprised of administering to a mammalian patient in

need of such treatment a compound of formula I or a pharmaceutically acceptable salt hydrate or pro-drug thereof in an amount which is effective for treating said ocular disease.

More particularly, a method of treating or preventing retinal vascularization is included herein, which is comprised of administering to a mammalian patient in need of such treatment a compound of formula I or a pharmaceutically acceptable salt, hydrate or pro-drug thereof in an amount which is effective for treating retinal vascularization. Diabetic retinopathy is an example of a disease in which neoangiogenesis or retinal vascularization is part of the overall disease etiology. Also included is a method of treating or preventing age-related macular degeneration.

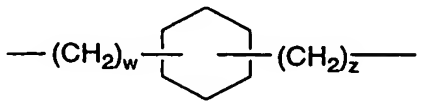
These and other aspects of the invention will be apparent from the teachings contained herein.

DETAILED DESCRIPTION OF THE INVENTION

The invention is described herein in detail using the terms defined below unless otherwise specified.

The term "alkyl" refers to a monovalent alkane (hydrocarbon) derived radical containing from 1 to 10 carbon atoms unless otherwise defined. It may be straight, branched or cyclic. Preferred straight or branched alkyl groups include methyl, ethyl, propyl, isopropyl, butyl and t-butyl. Preferred cycloalkyl groups include cyclopropyl, cyclobutyl, cycloheptyl, cyclopentyl and cyclohexyl.

Alkyl also includes a straight or branched alkyl group which contains or is interrupted by a cycloalkylene portion. Examples include the following:

and


wherein: x plus y = from 0-10; and w plus z = from 0-9.

The alkylene and monovalent alkyl portion(s) of the alkyl group can be attached at any available point of attachment to the cycloalkylene portion.

When substituted alkyl is present, this refers to a straight, branched or cyclic alkyl group as defined above, substituted with 1-3 groups of R^a, described herein.

The term "alkenyl" refers to a hydrocarbon radical straight, branched or cyclic containing from 2 to 10 carbon atoms and at least one carbon to carbon double bond. Preferably one carbon to carbon double bond is present, and up to four non-aromatic (non-resonating) carbon-carbon double bonds may be present. Preferred alkenyl groups include ethenyl, propenyl, butenyl and cyclohexenyl. As described above with respect to alkyl, the straight, branched or cyclic portion of the alkenyl group may contain double bonds and may be substituted with one to three groups of R^a, when a substituted alkenyl group is provided.

The term "alkynyl" refers to a hydrocarbon radical straight, branched or cyclic, containing from 2 to 10 carbon atoms and at least one carbon to carbon triple bond. Up to three carbon-carbon triple bonds may be present. Preferred alkynyl groups include ethynyl, propynyl and butynyl. As described above with respect to alkyl, the straight, branched or cyclic portion of the alkynyl group may contain triple bonds and may be substituted with 1-3 groups of R^a, when a substituted alkynyl group is provided.

Aryl refers to 5-10 membered aromatic rings e.g., phenyl, substituted phenyl and like groups as well as rings which are fused, e.g., naphthyl and the like. Aryl thus contains at least one ring having at least 5 atoms, with up to two such rings being present, containing up to 10 atoms therein, with alternating (resonating) double bonds between adjacent carbon atoms. The preferred aryl groups are phenyl and naphthyl. Aryl groups may likewise be substituted with 1-3 groups of R^a as defined herein. Preferred substituted aryls include phenyl and naphthyl substituted with one or two groups.

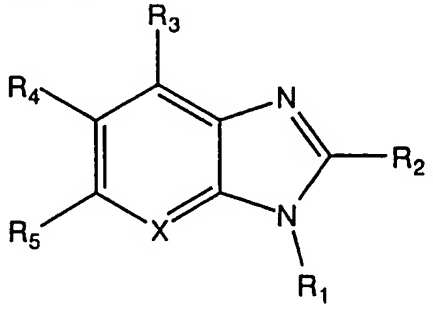
The term heterocycle, heteroaryl or heterocyclic, as used herein except where noted, represents a stable 5- to 7-membered mono- or bicyclic or stable 7- to 10-membered bicyclic heterocyclic ring system, any ring of which may be saturated or unsaturated, and which consists of carbon atoms and from one to three heteroatoms selected from the group consisting of N, O and S, and wherein the nitrogen and sulfur heteroatoms may optionally be oxidized, and the nitrogen heteroatom may optionally be quaternized, and including any bicyclic group in which any of the above-defined heterocyclic rings is fused to a benzene ring. The heterocyclic ring may be attached at any heteroatom or carbon atom which results in the creation of a stable structure. The heterocycle, heteroaryl or heterocyclic may be substituted with 1-3 groups of R^a. Examples of such heterocyclic elements include piperidinyl, piperazinyl, 2-oxopiperazinyl, 2-oxopiperidinyl, 2-oxopyrrolodinyl, 2-oxoazepinyl, azepinyl, pyrrolyl, 4-piperidonyl, pyrrolidinyl, pyrazolyl, pyrazolidinyl, imidazolyl, imidazolinyl, imidazolidinyl, pyridyl, pyrazinyl, pyrimidinyl, pyridazinyl, oxazolyl, oxazolidinyl, isoxazolyl, isoxazolidinyl, morpholinyl, thiazolyl, thiazolidinyl, isothiazolyl, quinuclidinyl, isothiazolidinyl, indolyl, quinolinyl, isoquinolinyl, benzimidazolyl, thiadiazoyl, benzopyranyl, benzothiazolyl, benzoxazolyl, furyl, tetrahydrofuryl, tetrahydropyranyl, thiophenyl, imidazopyridinyl, tetrazolyl, triazinyl, thienyl, benzothienyl, thiamorpholinyl sulfoxide, thiamorpholinyl sulfone, and oxadiazolyl. The term "alkoxy" refers to those groups of the designated length in either a straight or branched configuration and if two or more carbon atoms in length, they may include a double or a triple bond. Exemplary of such alkoxy groups are methoxy, ethoxy, propoxy, isopropoxy, butoxy, isobutoxy, tertiary butoxy, pentoxy, isopentoxy, hexoxy, isohexoxy allyloxy, propargyloxy, and the like.

The term "halogen" is intended to include the halogen atom fluorine, chlorine, bromine and iodine.

The term "prodrug" refers to compounds which are drug precursors which, following administration and absorption, release the drug in vivo via some metabolic process. Exemplary prodrugs include acyl amides of the amino compounds of this inventon such as amides of alkanoic(C_{1-6})acids, amides of aryl acids (e.g., benzoic acid) and alkane(C_{1-6})dioic acids.

Tyrosine kinase dependent diseases or conditions refers to hyperproliferative disorders which are initiated/maintained by aberrant tyrosine kinase enzyme activity. Examples include psoriasis, cancer, immunoregulation (graft rejection), atherosclerosis, rheumatoid arthritis, angiogenesis (e.g. tumor growth, diabetic retinopathy), etc.

The compounds of the present invention are in accordance with formula I:



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X is N or C;

R_1 is H, C_{1-10} alkyl, C_{3-6} cycloalkyl, C_{5-10} aryl, halo, OH, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl; said alkyl, alkenyl, alkynyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a;

R_2&R_3 are independently H, C_{1-6} alkyl, C_{5-10} aryl, C3-6 cycloalkyl, OH, NO_2, -NH_2, or halogen;

R_4 is H, C_{1-10} alkyl, C3-6 cycloalkyl, C_{1-6} alkoxy C_{2-10} alkenyl, C_{2-10} alkynyl, C_{5-10} aryl, C_{3-10} heterocyclyl, C_{1-6} alkoxyNR_7R_8, NO_2, OH, $-NH_2$ or C_{5-10} heteroaryl, said alkyl, alkenyl, alkynyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a;

R_5 is H, or C_{1-6} alkyl, OR, halo, NH_2 or NO_2;

R^a is H, C_{1-10} alkyl, halogen, NO_2, OR, $-NR$, NR_7R_8, R_7R_8, C_{5-10} aryl, C_{5-10} heteroaryl or C_{3-10} heterocyclyl,

R is H, or C_{1-6} alkyl, C_{1-6} alkylR_9;

R_9 is C_{5-10} aryl, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl; and

R7&R8 are independently H, C_{1-10} alkyl, C3-6 cycloalkyl, COR, C_{5-10} aryl, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl or NR7R8 can be taken together to form a heterocyclic 5-10 membered saturated or unsaturated ring containing, in addition to the nitrogen atom, one to two additional heteroatoms selected from the group consisting of N, O and S.

In one aspect, the invention is described wherein X is C and all other variables are as described above.

In another aspect, the invention is described wherein X is N and all other variables are as described above.

In still another aspect, the invention is described wherein R_4 is C_{1-10} alkyl, C_{3-6} cycloalkyl, C_{5-10} aryl, C_{5-10} heteroaryl, or C_{3-10} heterocyclyl, said alkyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a and all other variables are as described above.

In yet another aspect, the invention is described wherein R_1 is C_{1-10} alkyl, C_{5-10} aryl, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl, said alkyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a and all other variables are as described above.

A preferred subset of compounds of the present invention is realized when:

R_1 is H, C_{1-10} alkyl, C_{5-10} aryl, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl; said alkyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a;

R_2&R_3 are independently H, C_{1-6} alkyl, C_{3-6} cycloalkyl, OH, or halogen;

R_4 is H, C_{1-10} alkyl, C_{3-6} cycloalkyl, C_{5-10} aryl, C_{5-10} heteroaryl, C_{3-10} heterocyclyl, C_{1-6} alkoxyNR_7R_8, NO_2, OH, $-NH_2$ or said alkyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a; and all other variables are as described above.

Another preferred subset of compounds of the present invention is realized when:

R_1 is C_{5-10} aryl, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl; said aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a;

R_2&R_3 are independently H or C_{1-6} alkyl;

R_4 is C_{1-10} alkyl, C_{5-10} aryl, C_{5-10} heteroaryl, C_{3-10} heterocyclyl said alkyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one

to three members selected from R^a; and all other variables are as described above.

Examples of the compounds of this invention are:
1-phenyl-5-(4-methoxyphenyl)benzimidaole,
1-phenyl-5-(4-(2-(1-piperidinyl)ethoxy)phenyl)benzimidazole,
3-phenyl-6-(4-methoxylphenyl)imidazo[4,5-b]pyridine,
3-phenyl-6-(4-(2-(1-piperidinyl)ethoxy)phenyl)imidazo[4,5-b]pyridine,
3-phenyl-6-(4-(2-(1-piperidinyl)ethoxyphenyl)imidazo[4,5-b]pyridine,
3-(2-thiazoyl)-6-(4-(3-(1-piperidinyl)propylphenyl)imidazo[4,5-b]pyridine,
1-(2-thiazoyl)-5-(4-(3-(1-piperidinyl)propyl)phenyl)benzimidazol,
1-(3-thiophenyl)-5-(4-(3-(1-piperidinyl)propyl)phenyl)imidazo[4,5-b]pyridine,
1-(3-thiophenyl)-5-(4-(3-(1-piperidinyl)propyl)phenyl)benzimidazole,
3-(3-thiophenyl)-6-(4-(3-(1-piperidinyl)propylphenyl)imidazo[4,5-b]pyridine,
1-Phenyl-5-[5-(2-piperidin-1-yl-ethoxy)-pyridin-2-yl]-1H-benzimidazole,
1-(4-Cyanophenyl)-5-[6-(2-piperidin-1-yl-ethoxy)-pyridin-3-yl]-1H-benzimidazole,
1-Phenyl-5-[6-(2-piperidin-1-yl-ethoxy)-pyridin-3-yl]-1H-benzimidazole,
1-(3-Cyanophenyl)-5-[6-(2-piperidin-1-yl-ethoxy)-pyridin-3-yl]-1H-benzimidazole,
1-(3-Thiophene)-5-[6-(2-piperidin-1-yl-ethoxy)-pyridin-3-yl]-1H-benzimidazole,
[5-(1-Phenyl-1H-benzoimidazol-5-yl)-pyridin-2-yl]-(2-piperidin-1-yl-ethyl)-amine,
[5-(1-Phenyl-1H-benzoimidazol-5-yl)-pyridin-2-yl]-(2-morpholin-1-yl-ethyl)-amine,
4-(1-Phenyl-1H-benzoimidazol-5-yl)-1-(3-piperidin-1-yl-propyl)-1H-pyridin-2-one,
4-(1-Phenyl-1H-benzoimidazol-5-yl)-1-(3-piperidin-1-yl-ethyl)-1H-pyridin-2-one,
1-(3-Pyridyl)-5-(4-(2-(1-piperidinyl)ethoxy)phenyl)benzimidazole,
1-(4-Pyridyl)-5-(4-(2-(1-piperidinyl)ethoxy)phenyl)benzimidazole,
1-(3-Pyridyl)-5-[6-(2-piperidin-1-yl-ethoxy)-pyridin-3-yl]-1H-benzimidazole, and

1-(4-Pyridyl)-5-[6-(2-piperidin-1-yl-ethoxy)-pyridin-3-yl]-1H-benzimidazole or a pharmaceutically acceptable salt, hydrate or prodrug thereof.

The invention described herein includes a pharmaceutical composition which is comprised of a compound of formula I or a pharmaceutically acceptable salt or hydrate thereof in combination with a carrier. As used herein the terms "pharmaceutically acceptable salts" and "hydrates" refer to those salts and hydrated forms of the compound which would be apparent to the pharmaceutical chemist, i.e., those which favorably affect the physical or pharmacokinetic properties of the compound, such as solubility, palatability, absorption, distribution, metabolism and excretion. Other factors, more practical in nature, which are also important in the selection, are the cost of the raw materials, ease of crystallization, yield, stability, solubility, hygroscopicity and flowability of the resulting bulk drug.

When a compound of formula I is present as a salt or hydrate which is non-pharmaceutically acceptable, this can be converted to a salt or hydrate form which is pharmaceutically acceptable in accordance with the present invention.

When the compound is negatively charged, it is balanced by a counterion, e.g., an alkali metal cation such as sodium or potassium. Other suitable counterions include calcium, magnesium, zinc, ammonium, or alkylammonium cations such as tetramethylammonium, tetrabutylammonium, choline, triethylhydroammonium, meglumine, triethanolhydroammonium, etc. An appropriate number of counterions is associated with the molecule to maintain overall charge neutrality. Likewise when the compound is positively charged, e.g., protonated, an appropriate number of negatively charged counterions is present to maintain overall charge neutrality.

Pharmaceutically acceptable salts also include acid addition salts. Thus, the compound can be used in the form of

salts derived from inorganic or organic acids or bases. Examples include acetate, adipate, alginate, aspartate, benzoate, benzenesulfonate, bisulfate, butyrate, citrate, camphorate, camphorsulfonate, cyclopentanepropionate, digluconate, dodecylsulfate, ethanesulfonate, fumarate, glucoheptanoate, glycerophosphate, hemisulfate, heptanoate, hexanoate, hydrochloride, hydrobromide, hydroiodide, 2-hydroxyethanesulfonate, lactate, maleate, methanesulfonate, 2-naphthalenesulfonate, nicotinate, oxalate, pamoate, pectinate, persulfate, 3-phenylpropionate, picrate, pivalate, propionate, succinate, tartrate, thiocyanate, tosylate and undecanoate. Base salts include ammonium salts, alkali metal salts such as sodium and potassium salts, alkaline earth metal salts such as calcium and magnesium salts, salts with organic bases such as dicyclohexylamine salts, N-methyl-D-glucamine, and salts with amino acids such as arginine, lysine, and so forth. Also, the basic nitrogen-containing groups may be quaternized with such agents as lower alkyl halides, such as methyl, ethyl, propyl, and butyl chloride, bromides and iodides; dialkyl sulfates like dimethyl, diethyl, dibutyl; and diamyl sulfates, long chain halides such as decyl, lauryl, myristyl and stearyl chlorides, bromides and iodides, aralkyl halides like benzyl and phenethyl bromides and others. Other pharmaceutically acceptable salts include the sulfate salt ethanolate and sulfate salts.

The compounds of the present invention, may have asymmetric centers and occur as racemates, racemic mixtures and as individual diastereomers, or enantiomers with all isomeric forms being included in the present invention. When any variable (e.g., aryl, heterocyle, R1, etc)occurs more than one time in any constituent or in Formula I, its definition on each occcurence is independent of its definition at every other occurrence, unless otherwise stated.

The compounds of the invention can be formulated in a pharmaceutical composition by combining the compound with a

pharmaceutically acceptable carrier. Examples of such compositions and carriers are set forth below.

The compounds may be employed in powder or crystalline form, in solution or in suspension. They may be administered orally, parenterally (intravenously or intramuscularly), topically, transdermally or by inhalation.

Thus, the carrier employed may be, for example, either a solid or liquid. Examples of solid carriers include lactose, terra alba, sucrose, talc, gelatin, agar, pectin, acacia, magnesium stearate, stearic acid and the like. Examples of liquid carriers include syrup, peanut oil, olive oil, water and the like. Similarly, the carrier for oral use may include time delay material well known in the art, such as glyceryl monostearate or glyceryl distearate alone or with a wax.

Topical applications may be formulated in carriers such as hydrophobic or hydrophilic bases to form ointments, creams, lotions, in aqueous, oleaginous or alcoholic liquids to form paints or in dry diluents to form powders. Such topical formulations can be used to treat ocular diseases as well as inflammatory diseases such as rheumatoid arthritis, psoriasis, contact dermatitis, delayed hypersensitivity reactions and the like.

Examples of oral solid dosage forms include tablets, capsules, troches, lozenges and the like. The size of the dosage form will vary widely, but preferably will be from about 25 mg to about 500mg. Examples of oral liquid dosage forms include solutions, suspensions, syrups, emulsions, soft gelatin capsules and the like. Examples of injectable dosage forms include sterile injectable liquids, e.g., solutions, emulsions and suspensions. Examples of injectable solids would include powders which are reconstituted, dissolved or suspended in a liquid prior to injection.

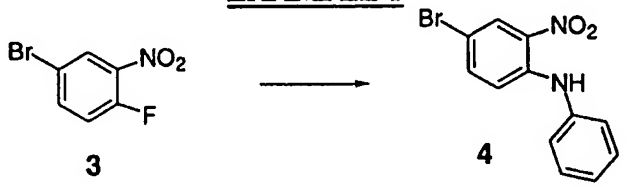
In injectable compositions, the carrier is typically comprised of sterile water, saline or another injectable liquid, e.g., peanut oil for intramuscular injections. Also, various buffering agents, preservatives and the like can be included.

For the methods of treatment disclosed herein, dosages can be varied depending upon the overall condition of the patient, the nature of the illness being treated and other factors. An example of a suitable oral dosage range is from about 0.1 to about 80 mg/kg per day, in single or divided doses. An example of a suitable parenteral dosage range is from about 0.1 to about 80 mg/kg per day, in single or divided dosages, administered by intravenous or intramuscular injection. An example of a topical dosage range is from about 0.1 mg to about 150 mg, applied externally from about one to four times a day. An example of an inhalation dosage range is from about 0.01 mg/kg to about 1 mg/kg per day.

The examples which follow illustrate the compounds that can be synthesized but they are not limited by the compounds in the tables nor by any particular substituents employed in the schemes for illustrative purposes.

The compounds may be administered in conventional dosages as a single agent or in combination with other therapeutically active compounds. The non-limiting examples that follow are illustrations of the compounds of the instant invention and are not meant to limit the invention in any way.

EXAMPLE 1

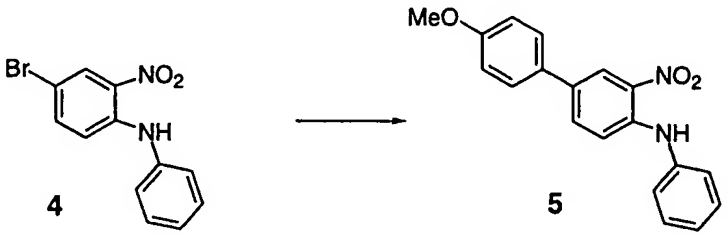


3 4

1-Bromo-4-fluoro-3-nitrobenzene (3) (1.14 mL, 9.06 mmol) was dissolved in 5 mL of anhydrous 1-methyl-2-pyrrolidinone under argon. Aniline (0.870 mL, 9.55 mmol) was added followed by the addition of *N,N*-diisopropylethylamine (1.90 mL, 10.9 mmol) and the resulting solution was heated to 120 °C. After 14 h additional aniline (0.082 mL, 0.90 mmol) was added and heating was continued for 8 h. The reaction solution was cooled to ambient temperature, diluted with

water and extracted with ethyl acetate (3x). The combined extracts was washed with brine, dried over Na_2SO_4, filtered and concentrated in vacuo to provide **4**.

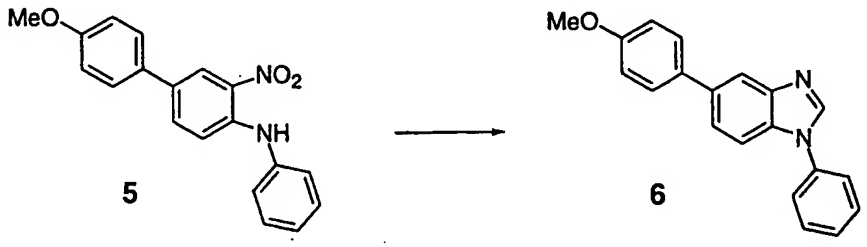
^{1}H NMR ($CDCl_3$) δ 9.46 (bs, 1H), 8.35 (d, 1H, J = 2.4 Hz), 7.45-7.40 (m, 3H), 7.29-7.25 (m, 3H), 7.10 (d, 1H, J = 9.2 Hz).



4 5

Bromoaromatic 4 (0.218 g, 0.744 mmol) and 4-methoxyboronic acid (0.125 g, 0.823 mmol) were dissolved in a mixture of dioxane (4 mL) and water (3 mL). Sodium carbonate (0.60 g, 5.7 mmol) was added and the resulting mixture was degassed and put under argon. Tetrakis(triphenylphosphine)palladium(0) (0.043 g, 0.037 mmol) was added and the reaction was heated to 80 °C. After 14 h the reaction was cooled to ambient temperature, diluted with water and extracted with ethyl actetate (3x). The combined extracts was dried with Na2SO4, filtered and concentrated to dryness. Purification by flash column chromatography (2 x 16 cm silica gel, 6:1 hexane/ethyl acetate) provided **5**.

^{1}H NMR (CDCl3) d 9.48 (bs, 1H), 8.40 (d, 1H, J = 2.4 Hz), 7.58 (dd, 1H, J = 2.4, 9.2 Hz), 7.50 (d, 2H, 9.0 Hz), 7.43 (t, 2H, J = 9.0 Hz), 7.32-7.22 (m, 4H), 6.98 (d, 2H, J = 9.0 Hz), 3.83 (s, 3H).

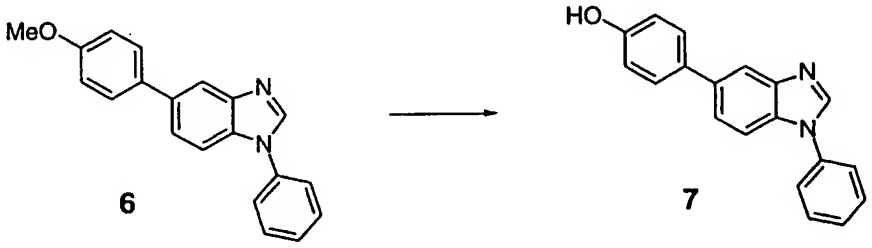


5 6

1-phenyl-5-(4-methoxyphenyl)benzimidaole.

Nitroaniline 5 (0.213 g, 0.665 mmol) and palladium on carbon (10%, 100 mg) were stirred in 8 mL 3:1 EtOH/AcOH. The reaction was put under a balloon of H_2. After 2 h the reaction was filtered through a plug of celite and the filtrate was concentrated to dryness. The resulting residue was dissolved in 1.5 mL trimethylorthoformate and heated to 120 ° C for 30 min. The solution was cooled concentrated to dryness and purified by flash column chromatography (2 x 15 cm silica gel, 1:1 hexane/ethyl acetate) which provided **6**.

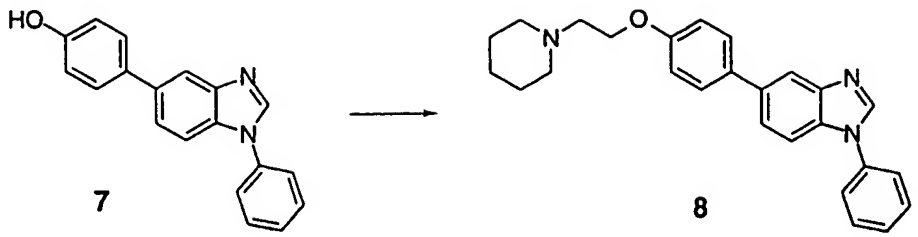
^{1}H NMR ($CDCl_3$) δ 8.14 (s, 1H), 8.04 (d, 1H, J = 0.9 Hz), 7.62-7.50 (m, 8H), 7.48 (t, 1H, J = 7.1 Hz), 7.01 (d, 2H, J = 8.8 Hz), 3.87 (s, 3H); FAB mass spectrometry $[M+H]^+$ 301.1; Anal. Calcd. for $C_{20}H_{16}N_2O$: C, 79.98; H, 5.37; N, 9.33. Found: C, 79.71; H, 5.48; N, 9.21.



6 **7**

An oven dried flask under argon was charged with benzimidazole **6** (0.039g, 0.13 mmol), aluminum chloride (0.175g, 1.31 mmol), and sodium iodide (0.200g, 1.33 mmol). Anhydrous acetonitrile (1 mL) and dichloromethane (0.5 mL) were added and reaction was heated to reflux. After 44 h the reaction was cooled to ambient temperature, quenched with water and extracted 3 x with ethyl acetate. The combined extracts was dried over Na_2SO_4, filtered and concentrated to dryness. The resulting residue was triturated with ether, filtered and dried to provide phenol 7.

^{1}H NMR ($CDCl_3$) δ 9.48 (s, 1H), 8.58 (s, 1H), 7.93 (s, 1H), 7.73-7.71 (m, 2H), 7.67-7.63 (m, 3H), 7.57-7.49 (m, 4H), 6.86 (d, 2H, J = 8.6 Hz).

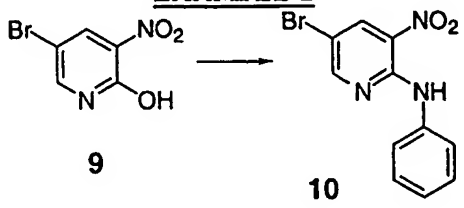


1-phenyl-5-(4-(2-(1-piperidinyl)ethoxy)phenyl)benzimidazole

Benzimidazole 7 (0.025g, 0.087 mmol) and N-(2-chloroethyl)piperidine hydrochloride (11 mg, 0.059 mmol) were dissolved in anhydrous N,N-dimethylformamide (0.5 mL). Cesuim carbonate (0.085g, 0.26 mmol) was added and the resulting mixture was heated to 50 °C. After 2 h additional and N-(2-chloroethyl)piperidine hydrochloride (11 mg, 0.059 mmol) was added. After 1 h the reaction was allowed to cool, quenched with water and extracted with ethyl acetate (3x). The combined extracts was washed with brine, dried over Na_2SO_4, filtered and concentrated to dryness. Purification by flash column chromatography (2 x 16 cm silica gel, 9:1 CH_2Cl_2/MeOH) provided **8** as a colorless oil.

^{1}H NMR ($CDCl_3$) δ 8.14 (s, 1H), 8.03 (d, 1H, J = 0.9 Hz), 7.62-7.50 (m, 8H), 7.48 (t, 1H, J = 7.2 Hz), 7.01 (d, 2H, J = 8.8 Hz), 4.21 (bt, 2H, J = 5.3 Hz), 2.87 (bs, 2H), 2.59 (bs, 4H), 1.66 (bs, 4H), 1.48 (bs, 2H); Mass spectrometry $[M+H]^+$ 398.3.

EXAMPLE 2

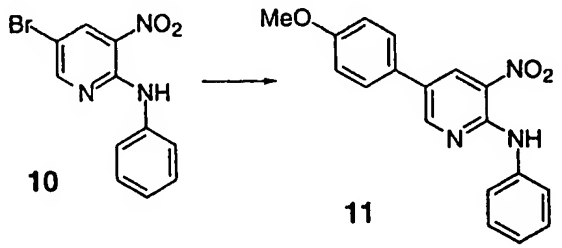


5-Bromo-2-hydroxy-3-nitropyridine (**9**) (5.736 g, 0.0262 mol) and 15mL thionyl chloride were added under argon. *N,N* dimethylformamide (1 mL) was then added and the solution was heated to reflux for 1 hr. By the end of the reaction, the

bromohydroxynitropyridine was completely dissolved in solution. After cooling to ambient temperature, 5 mL of toluene was added, and the solution was concentrated under vacuum. The product, 5-bromo-2-chloro-3-nitropyridine, was a yellow crystalline solid.

The bromochloronitropyridine was dissolved in 15mL of anhydrous 1-methyl-2-pyrrolidinone. Aniline (3.580 mL, 0.0393 mol) was added followed by the addition of *N,N* -diisopropylethylamine (13.69 mL, 0.0786 mol) and the solution was heated to 120 °C. After 1.5 hr., the solution was cooled to ambient temperature and diluted with water. The product was extracted using ethyl acetate and washed with brine. The organic layer was then dried over sodium sulfate, filtered, concentrated, and dried in vacuo. The crude mixture was purified using flash column chromatography (7.5 x 16 cm silica gel, 10:1 hexane:ethyl acetate) to afford **10** .

^{1}H NMR ($CDCl_3$) δ 10.04 (bs, 1H), 8.65 (dd, 1H, J = 2.2 Hz), 8.50(dd, 1H, J =2.4 Hz), 7.60 (d, 2H, J =8.6 Hz), 7.40(t, 2H, J =7.5 Hz), 7.21 (t, 1H, J =7.3 Hz).

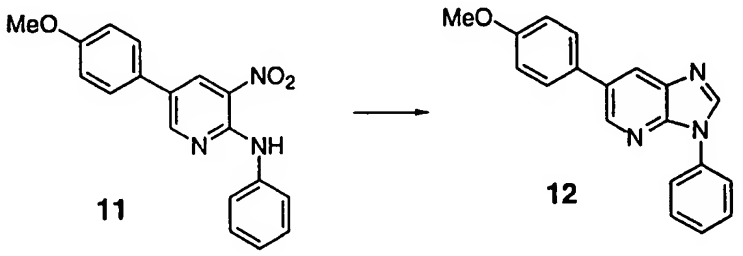


10 **11**

Bromoaromatic **10** (30 mg, 0.102 mmol), 4-methoxyphenylboronic acid (17 mg, 0.112 mmol) was dissolved in 0.75 mL dioxane followed by the addition of 204 µL of 2M sodium carbonate. The vessel was flushed with argon followed by the addition of tetrakis(triphenylphosphine)palladium(0) (6 mg, 0.005 mmol) and 0.56 mL water. The vessel was flushed again with argon and heated to 80 °C for 2.5 hr. The solution was cooled to room temperature and diluted with water. The product was extracted with ethyl acetate and washed with brine, followed by drying over sodium sulfate. The organic layer

was concentrated, and the product dried in vacuo. The crude mixture was purified by flash column chromatography (2.5 x 8 cm silica gel, 8:2 hexane:ethyl acetate), affording **11**.

^{1}H NMR ($CDCl_3$) δ 10.09 (bs, 1H), 8.71 (dd, 1H, *J* =2.4 Hz), 8.66 (dd, 1H, *J* =2.4), 7.67 (d, 2H, *J* =7.9), 7.49 (d, 2H, *J* =8.8 Hz), 7.41 (t, 2H, *J* =7.7 Hz), 7.18 (t, 1H, *J* =7.3 Hz), 7.00 (d, 2H, *J* =8.6 Hz), 3.86 (s, 3H).



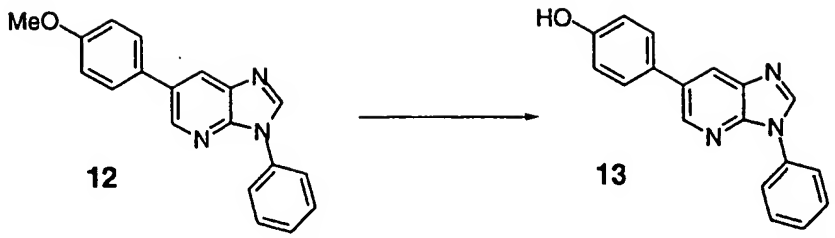
11 **12**

3-phenyl-6-(4-methoxylphenyl)imidazo[4,5-b]pyridine

Nitroaniline **11** (1.333 g, 4.15 mmol), Zn dust(6.239 g, 95.40 mmol), and 10 mL acetic acid were mixed under argon. The solution was heated to 60 °C for 1 hr until the solution turned light green. The zinc was removed using vacuum filtration with celite and washed with acetic acid. The filtrate was concentrated and 20 mL of trimethylorthoformate was added. The solution was heated to 100 °C for 2 hr followed by cooling to ambient temperature. The solution was concentrated and the crude mixture was purified by flash column chromatography(5 x 16 cm silica gel, 6:4 ethylacetate:hexane) affording **12**.

^{1}H NMR ($CDCl_3$) δ 8.61 (dd, 1H, *J* =2.0 Hz), 8.32 (s, 1H), 8.22 (dd, 1H, *J* =2.0), 7.74 (d, 2H, *J* =7.9 Hz), 7.55-7.50 (m, 4H), 7.39 (t, 1H, *J* =7.3), 6.99 (d, 2H, *J* =8.8 Hz), 3.80 (s, 3H). Mass spectrometry $[M+H]^+$ 302.3.

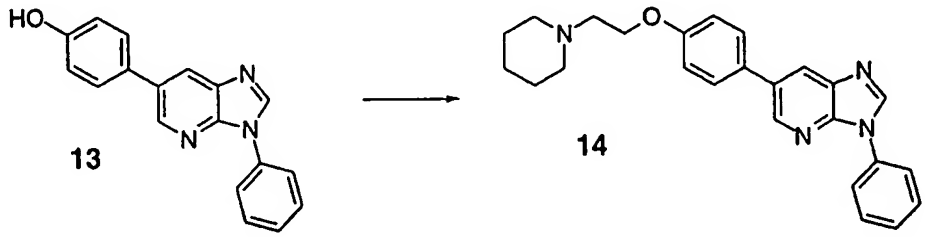
EXAMPLE 3



12 13

To the imidazopyridine **12** (202 mg, 0.670 mmol) was added a mixture of 10 mL hydrobromic acid and 10 mL acetic acid. The solution was stirred a room temperature for 5 min., followed by heating at 100 °C for 17 hr. The solution was cooled to ambient temperature and concentrated. Toluene (15 mL) was added and the solution was concentrated a second time. The concentrate was placed in vacuo over heating at 40 °C for 40 min., followed by further drying in vacuo at ambient temperature. Purification was acheived by reverse phase column chromatography affording **13**.

^{1}H NMR (CD_3OD) δ 9.45 (s, 1H), 8.82 (dd, 1H, *J* =1.8 Hz), 8.37 (dd, 1H, *J* =1.8 Hz), 7.91 (d, 2H, *J* =7.7 Hz), 7.68 (t, 2H, *J* =8.1 Hz), 7.63-7.57 (m, 3H), 6.95 (d, 2H, *J* =8.6 Hz).



13 14

3-phenyl-6-(4-(2-(1-piperidinyl)ethoxy)phenyl)imidazo[4,5-b]pyridine

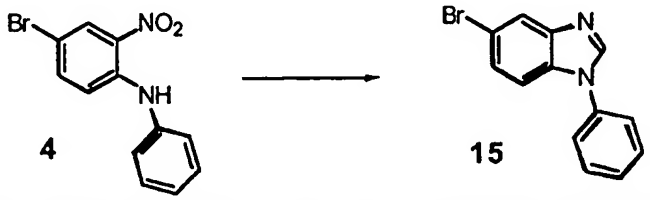
Cesium carbonate (296 mg, .908 mmol) and 1-(2-chloroethyl)piperidine monochlorohyrdide (84 mg, .454 mmol) were added under argon to a flame dried round bottom flask. Imidazopyridine **13** (87 mg, .303 mmol) was dissolved in 1.5 mL of anhydrous *N,N* dimethyl formamide under argon. The vessel was heated at 50 °C for 16 hr. and cooled to ambient temperature. The solution was diluted to 100mL with saturated sodium bicarbonate, and the product was extracted using ethyl acetate. The aqueous layer was extracted a

second time with dichloromethane w/ 3% 1-butanol. The organic layers were washed with saturated sodium bicarbonate, and dried over sodium sulfate. The organic layers were conentrated at aspirator pressure to remove ethyl acetate and methylene chloride; the 1-butanol and residual DMF were removed under high pressure. The product was purified using flash column chromatography(silica gel 2.5 x 32.5 cm, 10:1 methylene chloride:methanol). Excess trifluoroacetic acid was added to the product to create the resulting salt, and the mixture was triturated using ether. The TFA salt was dried using phosphorous pentoxide in vacuo to yield **14** (1.10 TFA salt).

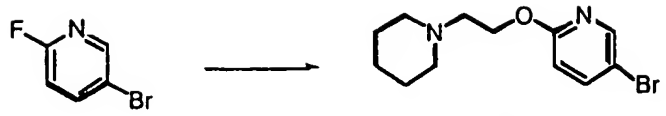
^{1}H NMR (CD_3OD) δ 8.66 (s, 1H), 8.55 (dd, 1H, J =2.0 Hz), 8.17 (dd, 1H, *J* =2.0 Hz), 7.82 (d, 2H, *J* =8.6 Hz), 7.59-7.52 (m, 4H), 7.46 (t, 1H, *J* =7.5 Hz), 7.01 (d, 2H, *J* =8.8 Hz), 4.85 (s, 2H), 4.15 (t, 2H, *J* =5.5 Hz), 2.84 (t, 2H, *J* =5.5 Hz), 2.62 (bs, 4H), 1.65 (m, 4H), 1.50 (m, 2H).

Anal. Calcd. for $C_{25}H_{26}N_4O$•1.10 TFA: C, 62.35; H, 5.21; N, 10.69. Found: C, 62.32; H, 4.93; N, 10.53.

EXAMPLE 4

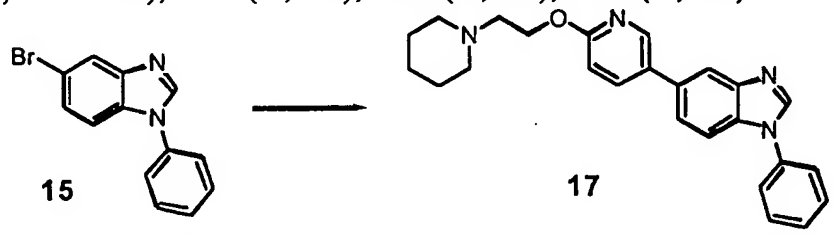


Bromoaromatic **4** (7.10 g, 24.1 mmol) and powdered zinc (36.2 g, 554 mmol, 23 equiv) were stirred in 80 mL glacial acetic acid. The mixture was heated to 60 °C. After 1h the reaction was cooled and filtered through a plug of celite and concentrated to dryness. The resulting residue was dissolved in 60 mL of formic acid and heated to 100 °C overnight. The reaction was cooled and concentrated to dryness. Purification by flash column chromatography (6x25 cm silica, 55:45 hexanes/EtOAc) afforded 5.88 g benzimidazole **15** (89% yield). ^{1}H NMR($CDCl_3$) δ 8.18 (s, 1H), 8.05 (d, 1H, *J*=1.7 Hz), 7.60 (t, 2H, *J*=7.1 Hz), 7.54-7.48 (m, 3H), 7.46 (dd, 1H, *J*= 1.8, 8.8 Hz), 7.40 (d, 1H, *J*=8.8 Hz).



16

1-Piperidineethanol (1.13 mL, 8.51 mmol) was dissolved in 10 mL anhydrous DMF under Ar. The solution was cooled to 0 °C and NaH (225 mg, 9.38 mmol) was added. After 10 min the mixture was allowed to warm to room temperature and 5-bromo-2-fluoropyridine (1.50 g, 8.52 mmol) was added. After 1h the reaction was quenched with water and extracted 3x with EtOAc. The combined extracts were dried over Na2SO4, filtered and concentrated to afford 2.20 g (91% yield) of the alkoxypyridine **16**. ^{1}H NMR($CDCl_3$) δ 8.17 (d, 1H, *J*=2.6 Hz), 7.62 (dd, 1H, *J*=2.6, 8.8 Hz), 6.67 (d, 1H, *J*=8.8 Hz), 4.40 (t, 2H, *J*=6.0 Hz), 2.74 (t, 2H, *J*=5.9 Hz), 2.49 (m, 4H), 1.60 (m, 4H), 1.44 (m, 2H).



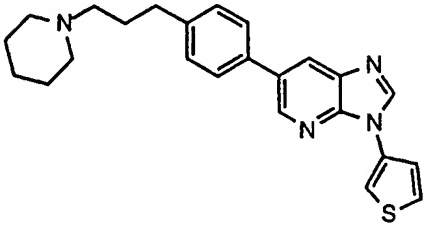
15 17

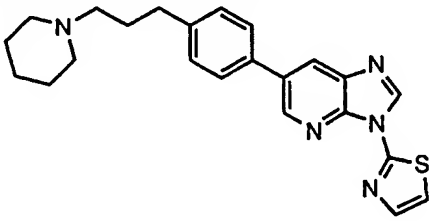
1-Phenyl-5-[6-(2-piperidin-1-yl-ethoxy)-pyridin-3-yl]-1H-benzimidazole

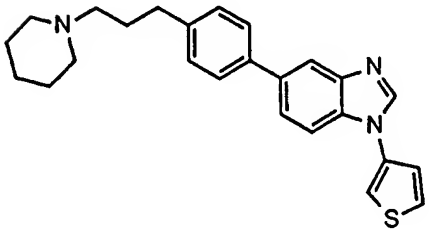
Benzimidazole **15** (2.91 g, 10.7 mmol), diboron pinacol ester (2.97 g, 11.7 mmol) and potassium acetate (3.14 g, 32.0 mmol) were stirred in 20 mL anhydrous DMF under Ar. $PdCl_2$(dppf) (0.26 g, 0.32 mmol) was added, solution was degassed and heated to 80 °C. After 20h the reaction was quenched with 125 mL of water and 50 mL of saturated aqueous NaCl and was extracted 3 x with EtOAc. The combined extracts were dried over Na_2SO_4, filtered and concentrated to afford 2.77 g of unpurified boronate. The unpurified boronate (650 mg, 2.03 mmol), alkoxypyridine **16** (526 mg, 1.85 mmol), 2M Na_2CO_3 (861 mg, 8.12 mmol), and 4 mL dioxane were added to a round bottom flask. After flushing three times with argon, $Pd(PPh_3)_4$ (117 mg, .10 mmol) was added, and the vessel was again flushed three times with argon. The vessel was heated to 80 °C under argon. After 22 hr., the reaction was cooled to room temperature followed by quenching with 25mL water. The mixture was extracted with 4x20 mL ethyl acetate, and the

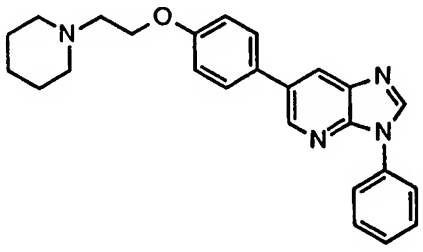
combined organic layers were washed with 1x20mL brine. The organic layer was dried over sodium sulfate, filtered, and concentrated. Purification was performed using reverse phase column chromatography (Waters 2x40mm C-18 column, H_2O:acetonitrile mobile phase gradient). The resulting oil was triturated with ether, filtered and washed with ether, affording **16**, a white TFA salt(150 mg, 16% yield). Mp: 160.5 – 162 °C. ^1H NMR($CDCl_3$) δ 8.41 (d, 1H, *J* =2.4 Hz), 8.19 (s, 1H), 8.01 (d, 1H, *J* =1.3 Hz), 7.90 (dd, 1H, *J* =11.0 Hz), 7.61 (m, 3H, *J* =13.6), 7.52 (m, 4H, *J* =31.0 Hz), 6.86 (d, 1H, *J* =8.4 Hz), 4.79 (t, 2H, *J* =9.9 Hz), 3.76 (bd, 2H, *J* =11.9 Hz), 3.51 (t, 2H, *J* =9.7 Hz), 2.80 (bt, 2H, *J* =23.1 Hz), 2.06 (m, 2H, *J* =26.2 Hz), 1.89 (s, 2H), 1.65 (s, 2H).

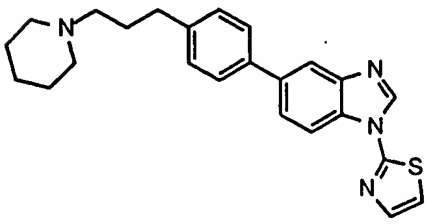
The following compounds can be made by literature methods and/or in combination with methods disclosed herein.











Kinase inhibition is demonstrated in accordance with the following protocol.

VEGF RECEPTOR KINASE ASSAY

VEGF receptor kinase activity is measured by incorporation of radio-labeled phosphate into polyglutamic acid, tyrosine, 4:1 (pEY) substrate. The phosphorylated pEY product is trapped onto a filter membrane and the incoporation of radio-labeled phosphate quantified by scintillation counting.

MATERIALS

VEGF receptor kinase

The intracellular tyrosine kinase domains of human KDR (Terman, B.I. et al. Oncogene (1991) vol. 6, pp. 1677-1683.) and Flt-1 (Shibuya, M. et al. Oncogene (1990) vol. 5, pp. 519-524) were cloned as glutathione S-transferase (GST) gene fusion proteins. This was accomplished by cloning the cytoplasmic domain of the KDR kinase as an in frame fusion at the carboxy terminus of the GST gene. Soluble recombinant GST-kinase domain fusion proteins were expressed in Spodoptera frugiperda (Sf21) insect cells (Invitrogen) using a baculovirus expression vector (pAcG2T, Pharmingen).

Lysis buffer

50 mM Tris pH 7.4, 0.5 M NaCl, 5 mM DTT, 1 mM EDTA, 0.5% triton X-100, 10 % glycerol, 10 mg/ml of each leupeptin, pepstatin and aprotinin and 1mM phenylmethylsulfonyl fluoride (all Sigma).

Wash buffer

50 mM Tris pH 7.4, 0.5 M NaCl, 5 mM DTT, 1 mM EDTA, 0.05% triton X-100, 10 % glycerol, 10 mg/ml of each leupeptin, pepstatin and aprotinin and 1mM phenylmethylsulfonyl fluoride.

Dialysis buffer

50 mM Tris pH 7.4, 0.5 M NaCl, 5 mM DTT, 1 mM EDTA, 0.05% triton X-100, 50 % glycerol, 10 mg/ml of each leupeptin, pepstatin and aprotinin and 1mM phenylmethylsuflonyl fluoride

10 X reaction buffer

200 mM Tris, pH 7.4, 1.0 M NaCl, 50 mM $MnCl_2$, 10 mM DTT and 5 mg/ml bovine serum albumin (Sigma).

Enzyme dilution buffer

50 mM Tris, pH 7.4, 0.1 M NaCl, 1 mM DTT, 10 % glycerol, 100 mg/ml BSA.

10 X Substrate

750 µg/ml poly (glutamic acid, tyrosine; 4:1) (Sigma).

Stop solution

30% trichloroacetic acid, 0.2 M sodium pyrophosphate (both Fisher).

Wash solution

15% trichloroacetic acid, 0.2 M sodium pyrophosphate.

Filter plates

Millipore #MAFC NOB, GF/C glass fiber 96 well plate.

METHOD

A. Protein purification

1. Sf21 cells were infected with recombinant virus at a multiplicity of infection of 5 virus particles/ cell and grown at 27 °C for 48 hours.

2. All steps were performed at 4°C. Infected cells were harvested by centrifugation at 1000 X g and lysed at 4 °C for 30 minutes with 1/10 volume of lysis buffer followed by centrifugation at 100,000Xg for 1 hour. The supernatant was then passed over a glutathione Sepharose column (Pharmacia) equilibrated in lysis buffer and washed with 5 volumes of the same buffer followed by 5 volumes of wash buffer. Recombinant GST-KDR protein was eluted with wash buffer/10 mM reduced glutathione (Sigma) and dialyzed against dialysis buffer.

B. VEGF receptor kinase assay

1. Add 5 µl of inhibitor or control to the assay in 50% DMSO.

2. Add 35 µl of reaction mix containing 5 µl of 10 X reaction buffer, 5 µl 25 mM ATP/10 µCi [^{33}P]ATP (Amersham), and 5 µl 10 X substrate.

3. Start the reaction by the addition of 10 µl of KDR (25 nM) in enzyme dilution buffer.

4. Mix and incubate at room temperature for 15 minutes.

5. Stop by the addition of 50 µl stop solution.

6. Incubate for 15 minutes at 4°C.

7. Transfer a 90 µl aliquot to filter plate.

8. Aspirate and wash 3 times with wash solution.

9. Add 30 µl of scintillation cocktail, seal plate and count in a Wallac Microbeta scintillation counter.

Human Umbilical Vein Endothelial Cell Mitogenesis Assay

Expression of VEGF receptors that mediate mitogenic responses to the growth factor is largely restricted to vascular endothelial cells. Human umbilical vein endothelial cells (HUVECs) in culture proliferate in response to VEGF treatment and can be used as an assay system to quantify the effects of KDR kinase inhibitors on VEGF stimulation. In the assay described, quiescent

HUVEC monolayers are treated with vehicle or test compound 2 hours prior to addition of VEGF or basic fibroblast growth factor (bFGF). The mitogenic response to VEGF or bFGF is determined by measuring the incorporation of [^{3}H]thymidine into cellular DNA.

Materials

HUVECs

HUVECs frozen as primary culture isolates are obtained from Clonetics Corp. Cells are maintained in Endothelial Growth Medium (EGM; Clonetics) and are used for mitogenic assays at passages 3-7.

Culture Plates

NUNCLON 96-well polystyrene tissue culture plates (NUNC #167008).

Assay Medium

Dulbecco's modification of Eagle's medium containing 1 g/ml glucose (low-glucose DMEM; Mediatech) plus 10% (v/v) fetal bovine serum (Clonetics).

Test Compounds

Working stocks of test compounds are diluted serially in 100% dimethylsulfoxide (DMSO) to 400-fold greater than their desired final concentrations. Final dilutions to 1X concentration are made directly into Assay Medium immediately prior to addition to cells.

10X Growth factors

Solutions of human $VEGF_{165}$ (500 ng/ml; R&D Systems) and bFGF (10 ng/ml; R&D Systems) are prepared in Assay Medium.

10X [^{3}H]Thymidine

[Methyl-^{3}H]Thymidine (20 Ci/mmol; Dupont-NEN) is diluted to 80 uCi/ml in low-glucose DMEM.

Cell Wash Medium

Hank's balanced salt solution (Mediatech) containing 1 mg/ml bovine serum albumin (Boehringer-Mannheim).

Cell Lysis Solution

1 N NaOH, 2% (w/v) Na_2CO_3.

Method

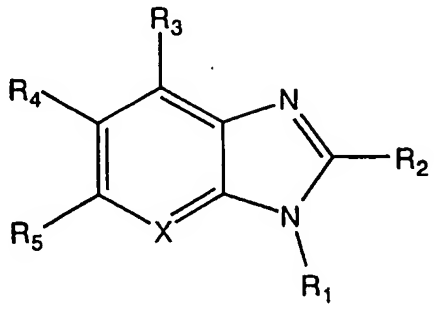
1. HUVEC monolayers maintained in EGM are harvested by trypsinization and plated at a density of 4000 cells per 100 ul Assay Medium per well in 96-well plates. Cells are growth-arrested for 24 hours at 37°C in a humidified atmosphere containing 5% CO_2.
2. Growth-arrest medium is replaced by 100 ul Assay Medium containing either vehicle (0.25% [v/v] DMSO) or the desired final concentration of test compound. All determinations are performed in triplicate. Cells are then incubated at 37°C/5% CO_2 for 2 hours to allow test compounds to enter cells.
3. After the 2-hour pretreatment period, cells are stimulated by addition of 10 ul/well of either Assay Medium, 10X VEGF solution or 10X bFGF solution. Cells are then incubated at 37°C/5% CO_2.
4. After 24 hours in the presence of growth factors, 10X [^{3}H]Thymidine (10 ul/well) is added.
5. Three days after addition of [^{3}H]thymidine, medium is removed by aspiration, and cells are washed twice with Cell Wash Medium (400 ul/well followed by 200 ul/well). The washed, adherent cells are then solubilized by addition of Cell Lysis Solution (100 ul/well) and warming to 37°C for 30 minutes. Cell lysates are

transferred to 7-ml glass scintillation vials containing 150 ul of water. Scintillation cocktail (5 ml/vial) is added, and cell-associated radioactivity is determined by liquid scintillation spectroscopy.

Based upon the foregoing assays the compounds of formula I are inhibitors of VEGF and thus are useful for the inhibition of neoangiogenesis, such as in the treatment of occular disease, e.g., diabetic retinopathy and in the treatment of cancers, e.g., solid tumors. The instant compounds inhibit VEGF-stimulated mitogenesis of human vascular endothelial cells in culture with IC_{50} values between 150-650 nM. These compounds also show selectivity over related tyrosine kinases (e.g. FGFR1 and the Src family).

WHAT IS CLAIMED IS:

1. A compound in accordance with formula I:



I

or a pharmaceutically acceptable salt, hydrate or prodrug thereof,

X is N or C;

R_1 is H, C_{1-10} alkyl, C_{3-6} cycloalkyl, C_{5-10} aryl, halo, OH, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl; said alkyl, alkenyl, alkynyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a;

R_2&R_3 are independently H, C_{1-6} alkyl, C_{5-10} aryl, C3-6 cycloalkyl, OH, NO_2, -NH_2, or halogen;

R_4 is H, C_{1-10} alkyl, C3-6 cycloalkyl, C_{1-6} alkoxy C_{2-10} alkenyl, C_{2-10} alkynyl, C_{5-10} aryl, C_{3-10} heterocyclyl, C_{1-6} alkoxyNR_7R_8, NO_2, OH, -NH_2 or C_{5-10} heteroaryl, said alkyl, alkenyl, alkynyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a;

R_5 is H, or C_{1-6} alkyl, OR, halo, NH_2 or NO_2;

R^a is H, C_{1-10} alkyl, halogen, NO_2, OR, -NR, NR_7R_8, R_7R_8, C_{5-10} aryl, C_{5-10} heteroaryl or C_{3-10} heterocyclyl,

R is H, or C_{1-6} alkyl, C_{1-6} alkylR_9;

R_9 is C_{5-10} aryl, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl; and

R7&R8 are independently H, C_{1-10} alkyl, C3-6 cycloalkyl, COR, C_{5-10} aryl, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl or NR7R8 can be taken together to form a heterocyclic 5-10 membered saturated or unsaturated ring containing, in addition to the nitrogen atom, one to two additional heteroatoms selected from the group consisting of N, O and S.

2. A compound in accordance with claim 1 wherein X is C and all other variables are as described above.

3. A compound in accordance with claim 1 wherein X is N and all other variables are as described above.

4. A compound in accordance with claim 1 wherein R_4 is C_{1-10} alkyl, C_{3-6} cycloalkyl, C_{5-10} aryl, C_{5-10} heteroaryl, or C_{3-10} heterocyclyl, said alkyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a and all other variables are as described above.

5. A compound in accordance with claim 1 wherein R_1 is C_{1-10} alkyl, C_{5-10} aryl, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl, said alkyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a and all other variables are as described above.

6. A compound in accordance with claim 1

wherein:

R_1 is H, C_{1-10} alkyl, C_{5-10} aryl, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl; said alkyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a;

R_2&R_3 are independently H, C_{1-6} alkyl, C_{3-6} cycloalkyl, OH, or halogen;

R_4 is H, C_{1-10} alkyl, C_{3-6} cycloalkyl, C_{5-10} aryl, C_{5-10} heteroaryl, C_{3-10} heterocyclyl, C_{1-6} alkoxyNR_7R_8, NO_2, OH, $-NH_2$ or said alkyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a; and all other variables are as described above.

7. A compound in accordance with claim 1

wherein:

R_1 is C_{5-10} aryl, C_{3-10} heterocyclyl, or C_{5-10} heteroaryl; said aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a;

R_2&R_3 are independently H or C_{1-6} alkyl;

R_4 is C_{1-10} alkyl, C_{5-10} aryl, C_{5-10} heteroaryl, C_{3-10} heterocyclyl said alkyl, aryl, heteroaryl and heterocyclyl being optionally substituted with from one to three members selected from R^a; and all other variables are as described above.

8. A compound in accordance with claim 1 which is:

1-phenyl-5-(4-methoxyphenyl)benzimidaole,
1-phenyl-5-(4-(2-(1-piperidinyl)ethoxy)phenyl)benzimidazole,
3-phenyl-6-(4-methoxylphenyl)imidazo[4,5-b]pyridine,
3-phenyl-6-(4-(2-(1-piperidinyl)ethoxy)phenyl)imidazo[4,5-b]pyridine,
3-phenyl-6-(4-(2-(1-piperidinyl)ethoxyphenyl)imidazo[4,5-b]pyridine,
3-(2-thiazoyl)-6-(4-(3-(1-piperidinyl)propylphenyl)imidazo[4,5-b]pyridine,
1-(2-thiazoyl)-5-(4-(3-(1-piperidinyl)propyl)phenyl)benzimidazol,
1-(3-thiophenyl)-5-(4-(3-(1-piperidinyl)propyl)phenyl)imidazo[4,5-b]pyridine,
1-(3-thiophenyl)-5-(4-(3-(1-piperidinyl)propyl)phenyl)benzimidazole,
3-(3-thiophenyl)-6-(4-(3-(1-piperidinyl)propylphenyl)imidazo[4,5-b]pyridine,
1-Phenyl-5-[5-(2-piperidin-1-yl-ethoxy)-pyridin-2-yl]-1H-benzimidazole,
1-(4-Cyanophenyl)-5-[6-(2-piperidin-1-yl-ethoxy)-pyridin-3-yl]-1H-benzimidazole,
1-Phenyl-5-[6-(2-piperidin-1-yl-ethoxy)-pyridin-3-yl]-1H-benzimidazole,
1-(3-Cyanophenyl)-5-[6-(2-piperidin-1-yl-ethoxy)-pyridin-3-yl]-1H-benzimidazole,
1-(3-Thiophene)-5-[6-(2-piperidin-1-yl-ethoxy)-pyridin-3-yl]-1H-benzimidazole,
[5-(1-Phenyl-1H-benzoimidazol-5-yl)-pyridin-2-yl]-(2-piperidin-1-yl-ethyl)-amine,
[5-(1-Phenyl-1H-benzoimidazol-5-yl)-pyridin-2-yl]-(2-morpholin-1-yl-ethyl)-amine,
4-(1-Phenyl-1H-benzoimidazol-5-yl)-1-(3-piperidin-1-yl-propyl)-1H-pyridin-2-one,
4-(1-Phenyl-1H-benzoimidazol-5-yl)-1-(3-piperidin-1-yl-ethyl)-1H-pyridin-2-one,
1-(3-Pyridyl)-5-(4-(2-(1-piperidinyl)ethoxy)phenyl)benzimidazole,
1-(4-Pyridyl)-5-(4-(2-(1-piperidinyl)ethoxy)phenyl)benzimidazole,
1-(3-Pyridyl)-5-[6-(2-piperidin-1-yl-ethoxy)-pyridin-3-yl]-1H-benzimidazole, and
1-(4-Pyridyl)-5-[6-(2-piperidin-1-yl-ethoxy)-pyridin-3-yl]-1H-benzimidazole or a pharmaceutically acceptable salt, hydrate or prodrug thereof.

9. A pharmaceutical composition which is comprised of a compound in accordance with claim 1 or a pharmaceutically acceptable salt, prodrug or hydrate thereof in combination with a carrier.

10. A method of treating or preventing cancer in a mammalian patient in need of such treatment which is comprised of admininstering to said patient an anti-cancer effective amount of a compound of claim 1.

11. A method of treating or preventing cancer in accordance with claim 8 wherein the cancer comprises cancers of the brain, genitourinary tract, lymphatic system, stomach, larynx and lung.

12. A method in accordance with claim 11 wherein the cancer comprises histiocytic lymphoma, lung adenocarcinoma, small cell lung cancers, pancreatic cancer, gioblastomas and breast carcinoma.

13. A method of treating or preventing a disease in which neoangiogenesis is implicated, which is comprised of administering to a mammalian patient in need of such treatment a compound of claim 1 or a pharmaceutically acceptable salt, prodrug or hydrate thereof in an amount which is effective for reducing neoangiogenesis.

14. A method in accordance with claim 13 wherein the disease is an ocular disease.

15. A method of treating or preventing retinal vascularization which is comprised of administering to a mammalian patient in need of such treatment a compound of claim

1 or a pharmaceutically acceptable salt, prodrug or hydrate thereof in an amount which is effective for treating retinal vascularization.

16. A method of treating or preventing diabetic retinopathy which is comprised of administering to a mammalian patient in need of such treatment a compound of claim 1 or a pharmaceutically acceptable salt, prodrug or hydrate thereof in an amount which is effective for treating diabetic retinopathy.

17. A method of treating or preventing age-related macular degeneration which is comprised of administering to a mammalian patient in need of such treatment a compound of claim 1 or a pharmaceutically acceptable salt, prodrug or hydrate thereof in an amount which is effective for inflammation.

18. A method of treating or preventing inflammatory diseases which is comprised of administering to a mammalian patient in need of such treatment a compound of claim 1 or a pharmaceutically acceptable salt, prodrug or hydrate thereof in an amount which is effective for inflammation.

19. A method according to claim 18 wherein the inflammatory disease comprises rheumatoid arthritis, psoriasis, contact dermatitis and delayed hypertensitivity reactions.

20. A method for inhibiting tyrosine kinase which comprises administering to a mammalian patient in need of such treatment a therapeutically effective amount of a composition of claim 1.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US98/19789

A. CLASSIFICATION OF SUBJECT MATTER
IPC(6) :C07D 235/08, 471/04; A61K 31/435, 31/415
US CL :548/304.4; 546/118, 193, 200; 514/394, 303
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED
Minimum documentation searched (classification system followed by classification symbols)
U.S. : 548/304.4; 546/118, 193, 200; 514/303, 394

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched
NONE

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
STN - FILE REGISTRY

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	Chemical Abstracts, Volume 78, Number 23, 23 April 1973, Stetsenko et al., "Imidacyanines From 2-methyl-5-phenyl-benzimidazole Derivatives", see page 68, column 1, abstract no. 112663k, Ref. Zh., Khim. 972, Abstract No. 9Zh 394. See compounds (I) and (II).	1, 2 and 4-7

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

*	Special categories of cited documents:	"T"	later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"A"	document defining the general state of the art which is not considered to be of particular relevance	"X"	document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"E"	earlier document published on or after the international filing date	"Y"	document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"L"	document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	"&"	document member of the same patent family
"O"	document referring to an oral disclosure, use, exhibition or other means		
"P"	document published prior to the international filing date but later than the priority date claimed		

Date of the actual completion of the international search
17 NOVEMBER 1998

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